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ATTRACTIVENESS TO MEXICAN FRUIT FLIES OF COMBINATIONS OF ACETIC ACID WITH AMMONIUM/AMINO ATTRACTANTS WITH EMPHASIS ON EFFECTS OF HUNGER

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Abstract—Ammonium acetate was more attractive than other ammonium salts to Mexican fruit flies (*Anastrepha ludens*) in an orchard test. We hypothesized that acetic acid enhanced the attractiveness of ammonia in the orchard test and that acetic acid may similarly enhance attractiveness of AMPu, an attractant consisting of a mixture of ammonium bicarbonate or ammonium carbonate, methylamine HCl, and putrescine. In laboratory experiments, acetic acid was attractive to flies deprived of either yeast hydrolysate or both sugar and yeast hydrolysate but not to flies fed both sugar and yeast hydrolysate. AMPu/acetic acid combinations were more attractive than AMPu alone to flies deprived of both sugar and yeast hydrolysate but not to flies fed sugar, regardless of yeast hydrolysate deprivation status. Acetic acid is the first attractant found that has become more attractive with both sugar and protein deprivation in studies with *A. ludens*. It is also the first that has enhanced the attractiveness of another attractant type. In orchard tests, yellow sticky panels baited with either AMPu or 17 mg of acetic acid were at least six times more attractive than unbaited panels. However, panels baited with both acetic acid (17–68 mg) and AMPu were less attractive than AMPu alone. These results differed from the laboratory data in which combinations were never less attractive than AMPu alone.

Key Words—Attractants, Mexican fruit fly, Diptera, Tephritidae, *Anastrepha ludens*, ammonia, methylamine, putrescine, acetic acid.

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INTRODUCTION

Ammonia has been used as a trap bait for Tephritidae since at least the 1920s (Ripley and Hepburn, 1929; Jarvis, 1931), predating even protein baits (McPhail, 1939; Steiner, 1952). Numerous ammonium salts have been used as sources of ammonia: ammonium sulfate (Prokopy and Economopoulos, 1975); ammonium bicarbonate (Haniotakis and Vassiliou-Waite, 1987); ammonium phosphate (monobasic) (Stavrakis, 1970); ammonium phosphate (dibasic), ammonium carbonate, ammonium oleate, and ammonium chloride (Gothilf and Levin, 1989); and ammonium acetate [Hodson (1943), Reissig (1974) and Prokopy (1975) with *Rhagoletis pomonella* Walsh; Burditt et al. (1983) with *Anastrepha suspensa* (Loew); Hedstrom and Jimenez (1988) with *A. obliqua* Macquart and *A. striata* Schiner; Gothilf and Levin (1989) and Heath et al. (1995) with *Ceratitis capitata* Wiedemann]. The great number of investigations using ammonium acetate as a fruit fly attractant testifies to its superior attractiveness relative to other ammonium salts.

Results of the above studies suggest that acetic acid, the form in which acetate volatilizes from solution, is the key to the superior attractiveness of ammonium acetate. Acetic acid has been reported attractive to *C. capitata* (Keiser et al., 1976) and to *Anastrepha ludens* Loew (Robacker and Flath, 1995). As discussed above, ammonia has long been acknowledged as an attractant for fruit flies. Therefore, we hypothesized that attractiveness of ammonium acetate probably was due to combined attractiveness of ammonia and acetic acid.

Robacker and Warfield (1993) described a three-component attractant (AMPu) for *A. ludens* that also utilizes ammonia as an attractive principal. AMPu consists of a mixture of ammonium bicarbonate or ammonium carbonate, methylamine HCl, and putrescine. AMPu was developed following the approach used by Wakabayashi and Cunningham (1991) to develop an attractant for *Bactrocera cucurbitae* Coquillett. The overall attractiveness of AMPu has been shown to be the result of additive effects of the three components.

In agreement with the above results, other studies in our laboratory have shown that combinations of attractive components within systems [for example, within the pheromone system (Robacker, 1988)] were more attractive than individual components to *A. ludens*. Conversely, combinations of chemicals from different systems (for example, pheromone with fruit odor) were either no more attractive or less attractive than the more attractive of the two systems (Robacker and Garcia, 1990). If ammonium acetate actually is more attractive than other ammonium salts, then we were interested in learning if AMPu/acetic acid combinations would also be more attractive than AMPu alone.

In addition to studies of component additivity on overall attractiveness, our laboratory has actively studied the effects of physiological state on responses of fruit flies to attractants in *A. ludens*. Most relevant to the current problem, effects

of hunger on attraction of flies to bacterial odor have been studied (Robacker and Garcia, 1993; Robacker and Moreno, 1995). Because bacterial odor contains acetic acid (Robacker and Flath, 1995), we wondered if hunger of flies would affect their attraction to acetic acid in the same way as to bacterial odor. The functional-group dissimilarity of acetic acid compared with the ammonia/amino compounds that comprise most of the attractive volatiles of bacterial odor suggested that acetic acid and bacterial odor would elicit different hunger-mediated responses.

The purposes of the current research were twofold: to determine the effects of sugar and protein hunger on attractiveness of acetic acid and AMPu to *A. ludens*, and to investigate interactions of acetic acid and AMPu with respect to their attractiveness, including effects of hunger on the interactions. The research was conducted as a series of three experimental paradigms. The first was a field test to verify that ammonium acetate was in fact more attractive than other ammonium salts to *A. ludens*. Next, we conducted laboratory experiments to determine effects of sugar and protein hunger on attraction of flies to AMPu, acetic acid, and their combination. Finally, field tests of AMPu, acetic acid, and their combination were conducted.

METHODS AND MATERIALS

Insects. A. ludens used in most experiments were from a culture that originated from mangoes collected in Morelos, Mexico, in 1953. Flies used in the orchard test of ammonium salts were from a culture that originated from yellow chapote fruit, a native host of the fly, collected in Nuevo Leon, Mexico, in 1987. Both cultures had been maintained on a laboratory diet since establishment. Flies used in orchard tests were irradiated with 70–84.7 Gy ^{137}Cs one to two days before adult eclosion for release into the orchard, to comply with quarantine laws for releasing *A. ludens*. Flies used in laboratory bioassays were not irradiated. Mixed-sex groups of 150–200 flies were kept in 0.5-liter cardboard cartons with screen tops until used in tests. Laboratory conditions for holding flies were 20–25°C, 50–70% relative humidity, and photophase from 06:30 to 19:30 hr provided by fluorescent lights.

Chemicals. AMPu originally was developed as an aqueous solution of ammonium bicarbonate, methylamine HCl, and putrescine in a 10:10:1 ratio (Robacker and Warfield, 1993). More recently, ammonium carbonate was substituted for ammonium bicarbonate because of its greater solubility in water, keeping the molar ratios of ammonia, methylamine, and putrescine the same as in the original AMPu (Robacker, 1995). Ammonium bicarbonate, methylamine HCl, and putrescine were obtained from Sigma Chemical Co. (St. Louis, Missouri), ammonium carbonate from Aldrich Chemical Co., Inc. (Milwaukee,

Wisconsin), and glacial acetic acid from Mallinckrodt Specialty Chemicals Co. (Paris, Kentucky). All chemicals were at least 98% pure.

Orchard Tests of Ammonium Salts (Experiment 1). A mixed citrus orchard located near the laboratory in Weslaco, Texas, was used for all field experiments. The orchard contained several varieties of orange, lemon, grapefruit and tangerine trees of varying ages. A section of the orchard containing Rio Red grapefruit (*Citrus paradisi* MacFadyen) was used for this experiment. Six treatments were tested in a 6 × 6 Latin-square design. Treatments were: 1% ammonium acetate; 5% ammonium phosphate (monobasic); 2.5% ammonium lactate; 2.5% ammonium sulfate; 2.5% ammonium citrate (dibasic); and 10% NuLure (Miller Chemical and Fertilizer Corp., Hanover, Pennsylvania) with 3% borax, a standard protein-hydrolysate bait for fruit flies. These concentrations of ammonium salts were more attractive than others tested in preliminary laboratory tests.

Treatments were prepared in deionized water. All except NuLure contained 0.01% Triton X-100 (Rohm and Haas Co., Philadelphia, Pennsylvania) as a surfactant. Treatments were tested in plastic McPhail-type traps, 200 ml of each per trap. Traps were hung one to a tree, 1–2 m aboveground, on the northeast sides of trees. Traps were placed in alternate trees within rows, and in alternate rows within the orchard. Positions of treatments were randomized within rows and columns. Flies were released into the orchard when 3–4 days old during the evening of the day before a test. Flies were fed a 6% sucrose solution up until the time of release. Approximately 5000 flies were distributed equally among the 36 test trees. Traps were placed in the orchard during the morning and removed for fly counts and cleaning two days later. The experiment was not repeated.

Laboratory Bioassays (Experiments 2–4). Three experiments were conducted to assess effects of sugar and protein hunger on attractiveness of AMPu, acetic acid, and their combination using cage-top bioassays. Attractiveness of acetic acid to Mexican fruit flies fed sugar but not protein was established in earlier work (Robacker and Flath, 1995). Of four quantities tested, 10 μg of acetic acid was the most attractive. Therefore, this quantity was used in the current research. AMPu was also tested at quantities of chemicals that were most attractive in previous work (Robacker and Warfield, 1993): ammonium bicarbonate, 10 μg; methylamine HCl, 10 μg; and putrescine, 1 μg. Both acetic acid and AMPu were prepared in water. The pH of the AMPu solution was adjusted to 8.8 with NaOH, a pH that was highly attractive in previous work (Robacker and Warfield, 1993). Test quantities of both acetic acid and AMPu were administered to flies in 10 μl of the aqueous solutions.

In the first laboratory experiment (experiment 2), flies fed ad libitum from eclosion on standard adult-fly diet, a mixture of sucrose (sugar from local grocery store) and enzymatic yeast hydrolysate (U.S. Biochemical Corp., Cleveland, Ohio). Additional sugar cubes were provided so that flies could regulate

the amount of carbohydrate and protein they received. In experiment 3, flies fed ad libitum on sugar cubes only. Experiment 4 was conducted like experiment 3 except sugar cubes were removed 48 hr before bioassays. Flies in all three experiments had access to water at all times.

Bioassays were conducted by placing four filter paper triangles (3 cm/side), one containing acetic acid, one with AMPu, one with acetic acid and AMPu, and one with water (10 μl), near the corners on the top of an insect cage (30 cm/side, aluminum-screened). For the papers containing both acetic acid and AMPu, the 10-μl quantities of each were loaded as separate spots so that non-volatile acetate salts did not form on the papers. Five counts of flies were made, at 1-min intervals, at each of the four treatments in their initial positions on the four corners of the cage-top. Then new papers with new loadings of the same four treatments were placed on the cage-top but rotated by 90° into new positions for five more counts, and so on, until all four treatments occupied each of the four positions for five counts. Totals of the 20 counts at each treatment were calculated, and these totals were used in statistical analyses. The filter papers were raised 5 mm above the cage top using plastic rings to ensure that olfaction and not contact chemoreception was solely responsible for the response of the flies.

One carton of 180–200 flies was used in each bioassay cage. Flies were tested when 5–9 days old. Tests were conducted between 10:00 and 14:00 hr under a combination of fluorescent and natural light. Sixteen replications of experiment 2, 14 replications of experiment 3, and 12 replications of experiment 4 were conducted.

Orchard Tests of AMPu and Acetic Acid (Experiments 5 and 6). One row of Ruby Red grapefruit (*C. paradisi*) and one row of Dancy tangerine (*C. reticulata* Blanco) were chosen for tests. Two linear blocks of eight consecutive trees each were used in each row, for a total of four blocks in the orchard.

AMPu and acetic acid were formulated into agar (Bacto Agar, Difco Laboratories, Detroit, Michigan) in 1.9-ml polypropylene microcentrifuge tubes (A. Daigger & Company, Inc., Wheeling, Illinois). Both the AMPu/agar tubes and the acetic acid/agar tubes were prepared by mixing hot agar solution with aqueous AMPu or acetic acid solutions in the microcentrifuge tubes to a volume of 1.7 ml. Final agar concentration was 1% in both AMPu and acetic acid tubes. AMPu concentrations in agar tubes were 60 mg/ml ammonium carbonate, 100 mg/ml methylamine HCl, and 10 mg/ml putrescine. Final pH of the AMPu tubes was 8.5–8.8. Acetic acid tubes were prepared at 10 concentrations of acetic acid, five of which were tested in experiment 5 and five in experiment 6. Different concentrations of acetic acid were tested in an attempt to test a range that included concentrations that were too low to have any effect up to concentrations that were so high as to be repellent. Concentrations tested in experiment 5 were 2.5, 5.0, 10, 20, and 40 mg/ml. Concentrations tested in experiment 6 were

0.06, 0.12, 0.25, 0.50, and 1.0 mg/ml. Experiment 6 used lower acetic acid concentrations because results of experiment 5 indicated that concentrations may have been too high.

Eight treatments were tested in each block. Experiment 5 treatments consisted of unbaited, AMPu, and acetic acid at 10 mg/ml and five combinations of the single concentration of AMPu and one of the five higher concentrations of acetic acid (2.5–40 mg/ml). Experiment 6 treatments consisted of unbaited, AMPu, and acetic acid at 0.25 mg/ml and five combinations of AMPu with one of the five lower concentrations of acetic acid (0.06–1.0 mg/ml). AMPu tubes and acetic acid tubes were fastened with their caps open to the tops of yellow panel traps (13 × 18 cm) (Robacker, 1992). For traps containing combinations of AMPu and acetic acid tubes, the two tubes were placed on opposite sides of the trap. Traps were coated with Tangle-Trap (Tanglefoot Company, Grand Rapids, Michigan) and were hung one to a tree, north of center, at 1–2 m height.

Flies were released into the test orchard when 4–14 days old during the late afternoon of the day before a test. Flies were fed sucrose and water until the time of release. Approximately 2000 flies were distributed equally among the 32 test trees in the four blocks. Traps were placed in the orchard during the morning and removed for fly counts and cleaning on the following morning. Traps were reused after removing flies and applying Tangle-Trap as necessary. Positions of treatments within each block were randomized for the first replication of each experiment. Positions of treatments in consecutive replications were not randomized but were moved sequentially within each block. Ten replications of experiment 5 and eight replications of experiment 6 were conducted.

Statistical Analyses. The field test of ammonium salts (experiment 1) was analyzed by analysis of variance (ANOVA) for a Latin-square design. Count data were analyzed after transformation to natural logarithms. Treatment means were compared by Fisher's protected least significant difference (LSD) method. These analyses and those described below were conducted using SuperANOVA (Abacus Concepts, 1989).

Two-way ANOVA was conducted for the three laboratory experiments (experiments 2–4) to separate effects of replication (each bioassay) and test chemical or chemical combination. Data used in ANOVAs were total counts at each treatment or water control for each bioassay resulting in counts of 16, 14, and 12 for experiments 2, 3, and 4, respectively. Means separations were by Fisher's protected LSD.

Separate one-way ANOVAs were conducted for males, females, and males plus females for each of the two field experiments of AMPu/acetic acid (experiments 5 and 6). Counts from traps were converted to proportions, then transformed by arcsin of the square root before ANOVAs were conducted. For example, the proportion of males captured in a particular trap on a particular

day was calculated as the number of males in that trap divided by the number of males captured in all traps in the block containing that trap on that day. Proportions were used to eliminate the high variability in capture rates from day to day because tests were conducted over two seasons. Means separations were by Fisher's protected LSD.

Sex ratios of flies captured by the lures were compared for experiments 5 and 6. Totals of females and males captured on each trap date by each lure type were used to calculate the proportion of females captured by each lure type per day. Data were analyzed by one-way ANOVA without transformation.

RESULTS

Orchard Tests of Ammonium Salts (Experiment 1). Ammonium acetate captured significantly more flies than the other ammonium salts (Figure 1). Only NuLure was comparable in attractiveness. A total of 1077 flies were captured in this experiment.

Laboratory Bioassays (Experiments 2–4). Acetic acid was no more attractive than water to flies fed both sugar and yeast hydrolysate until the time of testing (experiment 2) (Figure 2). Acetic acid was significantly more attractive than water to flies fed sugar but not protein (experiment 3) and to flies that were hungry for both sugar and protein (experiment 4). AMPu was significantly more attractive than both water and acetic acid to flies of all hunger-status groups.

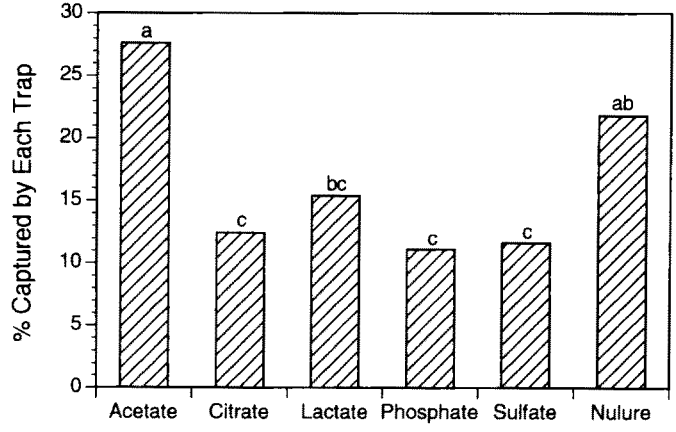

FIG. 1. Captures of *A. ludens* in McPhail-type traps containing solutions of various ammonium salts or NuLure (experiment 1), expressed as mean percentages of the total flies captured by the six treatments. Bars with the same letter are not significantly different by Fisher's protected LSD ($P < 0.05$).

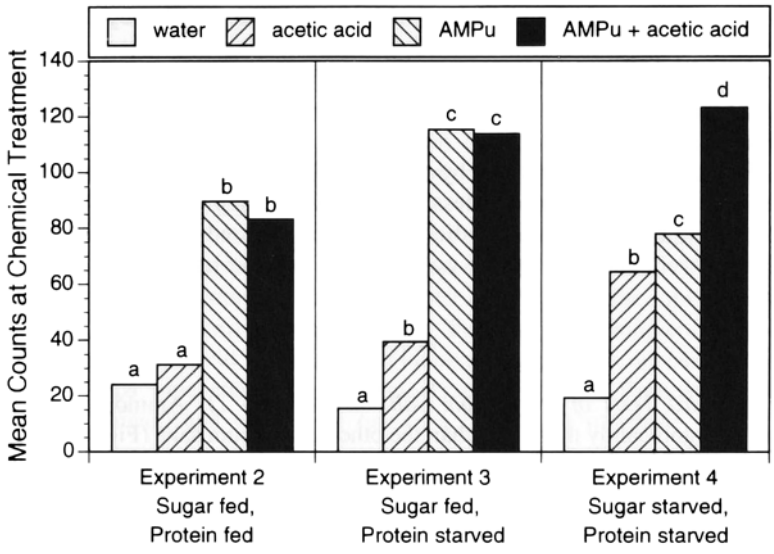

FIG. 2. Mean counts of *A. ludens* of three hunger states at filter papers containing water, acetic acid, AMPu, or an AMPu/acetic acid combination in cage-top bioassays. Within each experiment, bars with the same letter are not significantly different by Fisher's protected LSD ($P < 0.05$).

The combination of acetic acid and AMPu was more attractive than AMPu alone to flies hungry for both sugar and protein, but not to the other hunger-status groups of flies.

Orchard Tests of AMPu and Acetic Acid (Experiments 5 and 6). Experiment 5 showed that yellow panel traps baited with 17 mg (10 mg/ml × 1.7 ml) of acetic acid were considerably more attractive than unbaited yellow panel traps (Figure 3). AMPu-baited traps were not significantly more attractive than acetic acid-baited traps. Traps baited with combinations of acetic acid ranging in concentration from 2.5 to 40 mg/ml and AMPu were generally less attractive than traps baited with AMPu alone. The combinations were significantly less attractive than AMPu alone at the three highest concentrations of acetic acid. A total of 930 flies were captured in this experiment, summed over all treatments and replications.

In experiment 6, AMPu traps captured six times as many flies as unbaited traps, about the same as in experiment 5. The ANOVA was highly significant ($F = 16.2$; $df = 7,248$; $P < 0.0001$). Traps baited with 0.4 mg (0.25 mg/ml × 1.7 ml) of acetic acid were not significantly more attractive than unbaited

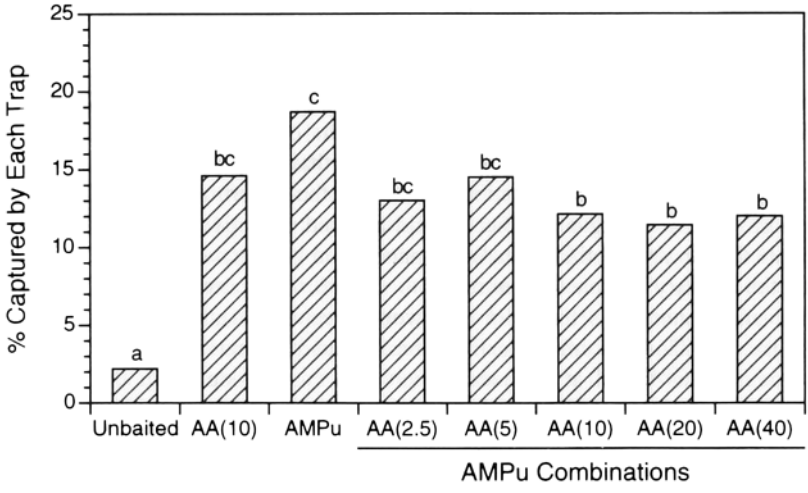

FIG. 3. Captures of *A. ludens* on sticky panel traps baited with nothing (unbaited), 17 mg of acetic acid [AA(10) = 10 mg/ml × 1.7 ml], AMPu, or combinations of AMPu with five amounts of acetic acid (experiment 5), expressed as mean percentages of the total flies captured by the eight treatments. Bars with the same letter are not significantly different by Fisher's protected LSD ($P < 0.05$).

traps by Fisher's protected LSD at the 5% level. Traps baited with combinations of acetic acid ranging in concentration from 0.06 to 1 mg/ml and AMPu also were not significantly different from traps baited with AMPu alone by LSD ($P < 0.05$). A total of 1640 flies were captured in this experiment, summed over all treatments and replications.

Most of the trap/lure combinations captured more females than males. The percentage of males and females in released flies was not recorded so it was not possible to determine if the various lures captured females or males in significantly different proportions than were present in the orchard. However, Robacker and Warfield (1993) showed that AMPu was approximately equally attractive to males and females.

We did compare the attractiveness to males vs females of the various lures relative to each other. In experiment 5, percentages of females captured by the various lures were unbaited, 52.4%; acetic acid, 49.6%; AMPu, 61.4%; and all AMPu/acetic acid combinations, 65.4%. In experiment 6, percentages of females captured by the lures were unbaited, 54.5%; acetic acid, 60.7%; AMPu, 57.6%; and all AMPu/acetic acid combinations, 59.5%. ANOVAs of proportions of females captured by traps per test day showed no significant differences among the lures.

DISCUSSION

Ammonium Acetate. The great attractiveness of ammonium acetate compared to the other ammonium salts suggests that acetic acid plays a role in attractiveness of ammonium acetate. The results of the experiments in which acetic acid was combined with AMPu proved that acetic acid affected overall attractiveness of these combinations. Based on the experiments with AMPu, it is more prudent to argue that acetic acid affected attractiveness of ammonium acetate than to argue otherwise. We cannot argue with certainty that acetic acid was the primary reason that ammonium acetate was more attractive than the other ammonium salts tested in this work because no attempt was made to equalize emissions of ammonia from the various preparations. However, each compound was tested at a concentration that ensured optimum performance based on preliminary testing.

Effects of Hunger on Attractiveness. The attractiveness of AMPu, acetic acid, and AMPu/acetic acid combinations was shown to be affected by the hunger status of flies. AMPu was attractive to flies of all hunger states. Although results of the experiments could not be compared statistically with each other, it appeared that AMPu was most attractive to flies hungry for protein (experiment 3) and least attractive to flies hungry for sugar (experiment 4). Previously, Robacker and Garcia (1993) demonstrated that bacterial odor was not attractive to *A. ludens* that were hungry for sugar, and Robacker and Moreno (1995) demonstrated that bacterial odor was most attractive to flies hungry for protein. The similar result found here for AMPu is not surprising due to the similarity of attractive principals in AMPu and bacterial odor (Robacker et al., 1993; Robacker and Flath, 1995).

Data for acetic acid revealed a very different relationship between hunger and attractiveness compared with AMPu. Acetic acid became more attractive as hunger for either sugar or protein increased. This relationship is also different from that of CEH, a three-component attractant (1,8-cineole, ethyl hexanoate, and hexanol) developed from odor of fermented host fruit (Robacker et al., 1990b). Like acetic acid, attractiveness of CEH increased with sugar deprivation of flies (Robacker, 1991). However, protein deprivation had little effect on attractiveness of CEH. Acetic acid is the first chemical studied that became more attractive to *A. ludens* deprived of either sugar or protein. With these data as a base, acetic acid does not fit into either the sugar-hunger or protein-hunger attractant systems described by Robacker (1991).

The greater attractiveness of the AMPu/acetic acid combination relative to either attractant alone when flies were starved for both sugar and protein is also unique among attractants studied to date in *A. ludens.* It is the first time that a combination of two attractants containing such different chemical moieties, and which operate somewhat differently with respect to effects of hunger, was more

attractive than one or the other of the two attractants alone. However, this result only occurred when flies were hungry for sugar and may occur in the field only at times when natural sources of carbohydrate are scarce.

Orchard Tests of AMPu/Acetic Acid Combinations. Combinations of AMPu with acetic acid were either less attractive or no more attractive than AMPu alone in field tests. This result cannot be attributed to acetic acid acting as a repellent. Acetic acid was attractive when tested alone, as discussed above. The field data resemble the laboratory data in that AMPu was the most attractive lure, acetic acid generally was more attractive than blanks, and the AMPu/acetic acid combinations were generally not more attractive than AMPu alone. In other ways, however, these results resemble neither laboratory experiments 2 and 3, in which acetic acid was much less attractive than AMPu and AMPu/acetic acid combinations, nor laboratory experiment 4, in which the AMPu/acetic acid combination was significantly more attractive than AMPu. The reason for these differences between the laboratory and field tests is unknown. Previous research indicated that irradiation attenuated attraction of *A. ludens* both to odor of fermented host fruit (Robacker et al., 1990a) and to bacterial odor (Robacker and Garcia, 1993) by similar amounts. What effects irradiation of flies used in field tests in this work may have had on relative attraction to acetic acid, AMPu, and their combinations cannot be predicted.

Diminution of attractiveness when attractants were combined has been reported before in *A. ludens.* Robacker and Garcia (1990) reported that the combination of attractive odor of fermented chapote fruit and male-produced pheromone was much less attractive to sexually mature virgin females than was pheromone alone. That work also showed that the combination was less attractive than chapote odor alone to sexually immature females. Robacker (1991) showed that a combination of CEH and bacterial odor was less attractive than CEH alone to 2- to 3-day-old flies that had not been fed as adults. In *B. oleae* (Gmelin), combinations of pheromone and proteinaceous feeding attractants were also found to be less attractive than the proteinaceous attractants alone (Haniotakis and Skyrianos, 1981; Zervas, 1989) or pheromone alone (Haniotakis and Vassiliou-Waite, 1987).

Zervas (1989) overcame the repellency problem of the pheromone/yeast hydrolysate combination by moving the pheromone dispenser 0.5–1 m away from the trap. This trapping system resulted in an increase in the capture of *B. oleae* compared to either bait alone. In the current study with *A. ludens,* putting AMPu vials and acetic acid vials on opposite sides of the panel traps did not result in an increase in the number of flies captured compared with AMPu alone. No attempt was made to duplicate Zervas' (1989) results by moving the AMPu and acetic acid lures 0.5–1 m apart.

As discussed above, Zervas (1989) successfully increased captures of *B. oleae* by combining two different types of attractants. In addition, Landolt et

al. (1992) demonstrated that a combination of pheromone from *Toxotrypana curvicauda* Gerstaecker with host-fruit odor was more attractive to females than either pheromone or host-fruit odor alone. Even more relevant to the current work, MacCollom et al. (1992, 1994) found that a combination of bacterial cells with apple volatiles was more attractive than either alone to *R. pomonella* in an apple orchard. These results indicate that it should be possible to develop more powerful attractants for at least some species of Tephritidae by combining different types of attractants. However, no evidence has been found to suggest that combining various types of attractants for the Mexican fruit fly, including AMPu with acetic acid, as shown in this paper, will enhance capture of the flies beyond captures by AMPu or other nitrogenous types of attractants alone.

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